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# Comparison of two methods for evaluating waste of a flow through trout farm

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## Abstract:

European water legislation enforces increasingly restrictive measures with regards to reduction of water consumption and waste emission in order to minimise the potential environmental impact of the agro industry sector. Fish farms are particularly concerned, but legislation covering effluent discharge varies significantly from country to country. However, recommendations and directives from institutional, national or regional bodies suggest the enforcement of increasingly strict waste reduction measures and the development of waste treatment. Before treatment, it is necessary to evaluate waste production in terms of composition and quantity. The waste quantification methods used today for fish culture systems are either based on direct measurements of nutrient and suspended solid fluxes or on indirect evaluation based on the digestibility coefficients of the feed constituents. The objective of the present study is to evaluate the waste of a freshwater flow through farm using both approaches and to discuss their applicability, drawbacks and advantages from the viewpoints of fish farmers and control authorities. Waste production on the farm was monitored during several 24 hour cycles in order to characterise the effluents of the system. The predictions and measurements for the total nitrogen (TN) parameter were well correlated, but measured and predicted suspended solids (SS) and total phosphorus (TP) values presented a weaker correlation coefficient. The hydrobiological method gives details on the N and P forms of waste but this method is heavy and it is difficult to obtain representative samples and flow rate measurements. The nutritional method is the simplest to use, provided that feed data are available.

**Keywords:** Waste; Trout farm; Suspended solids; Nitrogen; Phosphorus

## 25 **Introduction**

26 There are large differences in aquaculture regulations, in waste control and water quality  
27 survey methods and in legislation between European countries. In most countries, water  
28 quality is monitored by competent authorities and/or by self-monitoring (Fernandes *et al.*,  
29 2000; Bergheim and Brinker, 2003). Most countries have environmental quality standards  
30 mainly in relation to water quality and nutrient release. Some, such as Ireland or Norway,  
31 have brought in farming limitations based on a maximal stocking density or a maximal yearly  
32 feed quantity (Maroni, 2000). The aim of the EC Water Framework Directive (2000/60) is to  
33 develop a sustainable policy for environmental protection and especially, to homogenize all  
34 the directives or Community decisions adopted since 1975 on the fight against pollution and  
35 on the definition of water quality standards. Countries must progressively reduce polluted  
36 water emissions and develop monitoring programs with a view to improving water quality  
37 before 2015.

38 Concerning fish farm waste regulations, one may distinguish two different approaches: one  
39 based on a maximal authorized feed quantity; the other on maximal authorized emissions in  
40 the recipient ecosystem. In Denmark for example, the Danish decree (2002, November, 8<sup>th</sup>)  
41 fixed: (1) a maximal authorized annual feed quantity for freshwater farms, reduced or  
42 increased depending on water abundance and natural quantity and on the effluent treatment  
43 system, and (2) feed composition (energy, N, P and ash). A limit has been set on the tonnage  
44 of total nitrogen and phosphorous released into marine waters also (Pedersen, 1999). In  
45 France, the “polluter payer” principle implies that fish farmers must pay a tax to the regional  
46 water agencies. The payment is calculated on annual feed quantity and suspended solids (SS),  
47 nitrogen (N) and phosphorous (P) fluxes, with global emission coefficients obtained from  
48 feed digestibility determinations. Fish farm effluents are also regulated by the French ICPE

49 legislation (Classified Installations for the Protection of the Environment)<sup>1</sup>. This concerns  
50 fresh water farms and seawater farms with annual production above 10 metric tons and 20  
51 tons respectively. The key element of this legislation is the environmental impact assessment,  
52 in which waste quantification is required, and its impact evaluated. Therefore, in view of  
53 water legislation changes, fish waste characterisation and quantification are both key elements  
54 for fish farm operations and their waste monitoring and treatment.

55 For this purpose one may consider the particularities and origin of the wastes. Typically, fish  
56 waste is characterised by its high level of dilution when compared to other animal production  
57 or industrial wastewaters. The wastes first originate from the partial intake by fish or the  
58 partial digestibility of feed. When feed is metabolised by fish for energy and growth  
59 (including gamete production), as the efficiency of any biological reaction is less than 100%,  
60 some catabolites are produced in solid and soluble forms. Solid wastes, comprising faecal  
61 matter, constitute a more or less compact settleable material. Their chemical composition (C,  
62 N, P) and physical characteristics (size, density, water content...) depend on the feed  
63 composition and on the fish (species, phase of development). Large variations in nutrient  
64 utilisation by fish have been reported, depending on the type of nutrient (Kaushik, 1998). In  
65 addition to solids, faeces contain water and dissolved substances, mainly phosphorus and  
66 calcium. Fish also excrete soluble compounds through the gills and kidneys. When lipid and  
67 carbohydrate degradation produce CO<sub>2</sub> and water, protein degradation mainly produces  
68 ammonia (NH<sub>3</sub> and NH<sub>4</sub><sup>+</sup>), representing 80 to 90% of the soluble nitrogen excreted, with the  
69 balance being excreted mostly as urea. For most of the fish, nitrogen excretion represents 50  
70 to 70% of the nitrogen intake (Dosdat 1992a, b; Dosdat *et al.*, 1996; Company *et al.*, 1999).

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<sup>1</sup> Law No. 76-663 of July 19, 1976 with its decree of application No. 77-1133 of September 21, 1977. ICPE law has been codified in 2000 by the Environmental Code; the law is now abrogated and Book V Title 1<sup>st</sup> of the Environmental Code is the reference.

71 The main soluble phosphorus waste is orthophosphate ( $\text{PO}_4^{3-}$ ), representing only about 20%  
72 of the phosphorus intake (Dosdat *et al.*, 1996).

73 According to these characteristics, two different methods are used for fish culture systems: (1)  
74 a direct method, measuring dissolved and suspended matter in situ fluxes released by the  
75 farm, based on a hydrobiological approach and (2) an indirect evaluation, based on a  
76 nutritional approach, using defined feed amounts and appropriate digestibility coefficients  
77 (Jatteau, 1999).

78 In France, an expert panel<sup>2</sup> was appointed by the authorities to review the current strategies  
79 for evaluation of fish farm wastes (Papatriphon *et al.*, 2005). It was agreed that the method  
80 currently in use in France (Fauré, 1983) was not accurate enough and therefore should be  
81 replaced. This method uses the following equations to calculate waste production from  
82 salmonids :

83 (1)  $\text{NH}_4$  ( $\text{kg.d}^{-1}$ ) =  $K * \alpha * A$ , where  $K$  is a coefficient taking into account the  
84 number of previous water utilization ( $n$ ) with  $K = 0.8 + 0.2 * n$ ,  $A$  is the daily quantity of feed  
85 distributed ( $\text{kg.d}^{-1}$ ), and  $\alpha$  the  $\text{NH}_4$  production rate.

86 (2)  $\text{SS}$  ( $\text{kg.d}^{-1}$ ) =  $(1 - K_d) (33 * \text{IC} - 20) * A / 100$ , where  $K_d$  is the fish farm  
87 decantation coefficient and  $\text{IC}$  is the feed conversion ratio.

88 (3)  $\text{TP}$  ( $\text{kg.d}^{-1}$ ) =  $0.0048 * A$ .

89 The expert panel recommended a nutrient-balance model based on work by Cho *et al.* (1991),  
90 Cho and Bureau (1998) and Kaushik (1980, 1998). They carried out an initial validation of  
91 the model using data collected in 19 farms (self monitoring data and punctual measurements).  
92 This approach is based on the evaluation of the fish waste production through the digestibility

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<sup>2</sup> including scientists and representatives from (1) the French National Institute for Agricultural Research (INRA), the French Research Institute for the Exploitation of the Sea (IFREMER), (2) the feed manufacturing sectors, (3) the French Aquaculture Federation (FFA) and the Inter-Professional Committee of Aquaculture Products (CIPA)

93 of the distributed feed: waste production is given by the difference between the quantity of  
94 nutrient ingested and the part kept by the fish for its body gain.

95 The hydrobiological approach is based on the water flow rates and concentrations measured at  
96 the inlet and the outlet of the fish farm. Dissolved and particulate fluxes are calculated by  
97 subtracting the inlet flow from the outlet flow (Liao, 1970 and Liao and Mayo, 1974). Several  
98 studies were carried out (Fauré, 1983; Tarazona *et al.*, 1993, Kelly *et al.*, 1994; Lemarié *et al.*,  
99 1998), but the results were established for few fish species and feed compositions, while  
100 composition and digestibility coefficients change over time.

101 Boujard *et al.* (1999) compared the results of waste evaluation using the nutritional and  
102 hydrobiological approaches on several rainbow trout breeding tanks. Nutrient concentrations  
103 and flow rate measurements were carried out two times, during two consecutive days, with  
104 water sampling and flow rate measurements every two hours. Water sampling and flow rate  
105 measurements methods are not described in the publication. Good correlations between  
106 measured and predicted values were found, but they found that the predicted values were  
107 always underestimated. Papatryphon *et al.* (2005) compared values predicted by a nutrient-  
108 balance model with fluxes calculated from nutrient concentrations measured in the recipient  
109 river. The water flow rates and nutrient concentrations were not directly measured during the  
110 study but were collected from farmers or water agency records. They found a good  
111 correlation, but a tendency to overestimate the predicted  $\text{NH}_4^+$  and P values.

112 This approaches raise the problem of (1) synchronization between nutrient concentrations and  
113 flow rate measurements and (2) the accuracy of the water flow rates and nutrient  
114 concentration measurements, which are the two key elements to evaluate waste fluxes.

115 In this study, in order to optimise the accuracy on the mass balance evaluation, our approach  
116 consisted in simultaneous measurements of nutrient concentrations and flow rates, 4 times  
117 during 24h periods, using the same methodology and measurements devices located at the

118 same sampling spots. Continuous data acquisition equipment was used in order to optimise  
119 the precision of the measurements.

120 The first objective of our study was to compare the nutrient fluxes obtained using both current  
121 approaches in order to evaluate the waste produced by a whole flow through farm, with  
122 continuous sampling during several 24 hour periods in order to characterise the daily waste  
123 fluxes.

124 The second objective was to discuss the applicability of both waste evaluation approaches for  
125 the fish farmers and control authorities, as tools for the waste quantification which is required  
126 in the French ICPE legislation.

127

## 128 **Materials and methods**

129 The investigation took place in 2005-2006 on the on-growing unit of the Charles Murgat SA  
130 trout farm, located at Beaurepaire in south east France. The farm is operated using the flow  
131 through system and produces on average 600 tons of brook trout, brown trout, rainbow trout  
132 and arctic char per year, at a fish stocking density of around  $58 \text{ kg.m}^{-3}$ .

133 The on-growing unit is divided into two sectors (figure 1):

134 - sector 1 is composed of 7 concrete raceways (each  $70*6*0.8 \text{ m}$ ), with 4 species reared  
135 from 50 g to more than 2000 g. Each tank is divided into batches, each comprising  
136 different species, at different sizes, corresponding to the market demand. During the  
137 studied period, 55 to 70 % of the fish weighed around 200 g and the average feed  
138 conversion ratio (FCR) was 0.85.

139 - sector 2 is composed of 2 concrete raceways, with only rainbow trout species (from  
140 200 g to 1000 g.). The average weight of 50 % of the population is around 500 g and  
141 the average FCR is 0.95.

142 Both sectors use very high quality, constant temperature well water (around 10 °C during the  
143 period). The first three tanks of sector 1 are fed with a well water flow rate varying from 600  
144  $\text{l}\cdot\text{s}^{-1}$  up to 2000  $\text{l}\cdot\text{s}^{-1}$ , corresponding to a water renewal rate of between 200 % and 600 % per  
145 hour in the tanks. After a first use, the rearing water is filtered through a mechanical filter,  
146 oxygenated, and reused in the four following tanks of the sector 1. Each tank is equipped with  
147 several aerators in order to keep the oxygen concentration above 5  $\text{mg O}_2\cdot\text{l}^{-1}$  in the tank outlet.  
148 The effluent of that sector is filtered with another drum filter before being released into the  
149 river through a sport fishing area. The two tanks of sector 2 are fed with the same well water,  
150 with a flow rate varying around 500  $\text{L}\cdot\text{s}^{-1}$ .

151 In this study the wastes produced by the two on-growing units (sectors 1 and 2) of the farm  
152 were evaluated using the hydrobiological and the nutritional methods.

### 153 *The “hydrobiological” method*

154 The hydrobiological method is based on water sampling and flow rate measurements. In order  
155 to optimise the accuracy of the flow rate measurement, it was decided to measure the water  
156 velocity in the tanks which are easily accessible, have a well defined cross section) and a  
157 more homogeneous hydraulic regime than the water inlet and outlet channels. Four 24h  
158 sampling periods were performed on sectors 1 and 2 between January and March 2006, the  
159 last one only on sector 1 (sector 2 was not sampled because of important fishing events). The  
160 sampling period was fixed for 24h because the feeding ratio is stable over a period of two  
161 days. The inlet and the outlet waters of the two sectors were sampled by ISCO 6712  
162 automatic sampler over 24h, with a frequency of one sample every 30 minutes in order to  
163 follow the daily fluctuations of waste concentrations linked to the feeding periods (Hennessy  
164 et al, 1996). Water samples were stored 24 hours at 4°C before analysis. In water samples,  
165 dissolved N and P, particulate N and total P and suspended solids concentrations were  
166 measured.

167 Dissolved N and P were measured by spectrophotometry, after filtration on Whatman GF/C  
168 filters.  $\text{NH}_4\text{-N}$ ,  $\text{NO}_2\text{-N}$ , urea-N,  $\text{PO}_4\text{-P}$  were analysed using an Alliance Instruments Evolution  
169 II, after AFNOR method (NF T 90-015) described by Solorzano (1969) and the ISO method  
170 (6777-1984 F) described by Bendschneider and Robinson (1952) respectively.  $\text{NO}_3\text{-N}$  was  
171 measured with a Technicon® Autoanalyzer II, after a nitrite reduction on a cadmium-copper  
172 column (Wood *et al.*, 1967).

173 Particulate-N was obtained after a CHN analysis and total-P by using a colorimetric method  
174 NFENISO11885 (after mineralisation). Total N was calculated by adding the nitrogenous  
175 compound concentrations. Suspended solid (SS) concentrations were determined after GF/C  
176 filtration (NFEN872).

177 During the sampling periods, the water flow rates were measured with a bottom mounted  
178 Argonaut- shallow water Doppler current meter (Huhta and Ward, 2003). This current meter  
179 provides a vertically integrated velocity measurement (4 points of measurement in the water  
180 column). The water flow rates were measured in the 9 tanks of the farm, which constitute the  
181 two sectors, with a frequency of one sample every 15 seconds. The current meter was placed  
182 on the bottom of the tanks and moved at different distances of the vertical walls (every 50 cm)  
183 during the 24 hour period. These measures enabled calculation of the average water flow rate  
184 of the farm. The effluent (dissolved, particulate and SS) fluxes produced by the fishes during  
185 the 24 hour period were calculated by subtraction of inlet fluxes from outlet fluxes.

186 Temperature, oxygen, pH and redox were also controlled with a Consort multi-parameter  
187 analyser.

188

189 *The “nutritional” method*

190 Fish farm effluent production was calculated with the nutrient balance model developed by  
191 Papatryphon *et al.* (2005). This model is based on feed utilisation by the fish. Waste fluxes



192 are calculated by removing the part retained by the fish (biomass production and body  
193 composition), from the part ingested by the fish.

194 Total effluents include solid and dissolved effluents, with solid effluents as the undigested  
195 part of the feed (calculated with the nutrient digestibility coefficients (Guillaume *et al.*,  
196 1999)), and dissolved effluents as the rest. The total-SS are calculated by adding the faecal  
197 SS, equivalent to the non digested feed (proteins, lipids, carbohydrates, ash and fibres) and the  
198 SS from uneaten feed. In this method, the following equations are used to calculate N, P and  
199 SS waste production:

200 **(1) Total nitrogen = solid nitrogen + dissolved nitrogen**

201 **Solid N** = Faecal N + Uneaten N

202 Faecal N =  $[(DF - (DF * \% UF))] * (\% \text{ protein} / 6.25) * (100 - DC) \%$

203 Uneaten N =  $(DF * \% UF) * (\% \text{ protein} / 6.25)$

204 With: DF = distributed feed, UF = uneaten feed, % protein = proportion of protein in feed,  
205 DC = digestibility coefficient

206 **Dissolved N** = consumed N – faecal N – digested part of N

207 Consumed N =  $DF - (DF * \% UF) * (\% \text{ protein} / 6.25)$

208 Digested part N =  $DF * BN / FCR$

209 With BN = Whole fish body N content = 0.0256-0.0272 g/g of body weight (Papatriphon et  
210 al, 2005); FCR = Feed Conversion Ratio. The dissolved NH<sub>4</sub>N is calculated with an 80 %  
211 coefficient corresponding to the proportion of NH<sub>4</sub>N in total dissolved N excretion  
212 (Papatriphon et al, 2005).

213 Similar equations with appropriate coefficients are used to evaluate P wastes: the proportion  
214 of phosphorus in feed composition and the whole fish body P content of 0.004 g/g of body  
215 weight (Papatriphon et al, 2005).

216

217 (2) **Total SS = faecal SS + uneaten feed SS**

218 **Faecal SS** = Non digested proteins + Non digested lipids + Non digested  
219 carbohydrates + Non digested ash + Non digested fibres

220 
$$=[(DF - (DF * \% UF)] * \sum [\% \text{nutriment} \times (100-DC)\% ]$$

221 **Uneaten feed SS** = (DF \* % UF) \* (% dry matter in feed)

222 The digestibility coefficients (DC) were those proposed by Papatryphon et al., 2005 (table I);  
223 protein and lipid digestibility coefficients were compared to the digestibility coefficient  
224 measured by the manufacturer.

225 Fish were fed twice a day around 1 % of the standing stock per day, with two different feed  
226 origins according to the fish size. The average feed composition is presented on table I. Fish  
227 were fed partly automatically, partly manually, up to satiety. The daily feed quantity  
228 distributed manually was determined from feeding tables by a computerised distribution  
229 system. The complementary quantity distributed manually up to satiety was also registered.  
230 This feeding method allowed avoiding uneaten feed. Tank biomass was evaluated from the  
231 biometrics every other week (average weight on 50 fish, for each batch) and enabled  
232 calculation of the FCR. Body nutrient contents were set on 26 g N. kg<sup>-1</sup> of body weight and 4  
233 g P. kg<sup>-1</sup> of body weight (Papatryphon *et al.*, 2005).

234

## 235 **Results**

236 Daily feed rate and tank biomass were stable during the studied period. The biological data  
237 are presented in table II. The water flow rate of the whole farm fluctuated around 1336.7 ±  
238 210.8 l.s<sup>-1</sup> (average daily flow rates of 820, 840, 1030 and 857 l.s<sup>-1</sup> on sector 1, during the four  
239 24 h periods respectively, and 400, 370 and 550 l.s<sup>-1</sup> on sector 2, during the three 24 h  
240 periods). 168 samples were treated.

241 The daily waste fluxes of the farm, predicted with the nutritional method, the CEMAGREF  
242 method and measured with the hydrobiological method are presented in table III, with  
243 corresponding values expressed as fluxes per kg feed. These data correspond to the waste  
244 produced by a standing stock of 132 tonnes of fish (average value during the studied period).  
245 The daily average flux of total-N, measured using the hydrobiological method is  $54.1 \pm 10$   
246  $\text{kg.d}^{-1}$ , when the predicted value is  $59.82 \pm 6.01 \text{ kg.d}^{-1}$ . The measured daily flux of total-P is  
247  $13.6 \pm 3.5 \text{ kg.d}^{-1}$ , almost twice the predicted value:  $6.33 \pm 0.61 \text{ kg.d}^{-1}$ . The measured daily  
248 flux of SS is  $317.8 \pm 165.7 \text{ kg.d}^{-1}$  compared to a predicted value of  $206.48 \pm 20.67 \text{ kg.d}^{-1}$ . The  
249 measured fluxes of particulate-N,  $\text{NH}_4\text{-N}$  and urea-N are respectively  $11.8 \pm 3.4$ ,  $31.6 \pm 7.5$   
250 and  $10.7 \pm 2.5 \text{ kg.d}^{-1}$  and the particulate-P and  $\text{PO}_4\text{-P}$  fluxes produced by the fish are  $9.6 \pm 3.6$   
251 and  $4.0 \pm 0.2 \text{ kg.d}^{-1}$  (table III).

252 Using the CEMAGREF method (Fauré, 1983),  $\text{NH}_4\text{-N}$ , TP and SS fluxes of the farm are  $36.4$   
253  $\pm 3.7$ ,  $6.7 \pm 0.7$ , and  $136.3 \pm 14.1 \text{ kg.d}^{-1}$  respectively (table III).

254 Variance of the predicted and measured fluxes represents the variability of the fluxes between  
255 each 24 hour period. The figure 2 presents a comparison between predicted and measured  
256 fluxes.

257 Figures 3 - 5 show the relation between measured and predicted TN, TP and SS. The  
258 measured and predicted TN values are well correlated with  $r^2 = 0.88$ ) whereas the correlation  
259 coefficients between measured and predicted TP and SS values are weaker (0.53 and 0.48  
260 respectively).

261 The hydrobiological method provides detailed information on the different forms of nitrogen  
262 and phosphorous fluxes; 21% of nitrogen wastes are in the particulate form, 59% are  $\text{NH}_4\text{-N}$   
263 and 20% urea-N. 68.8% of the phosphorous wastes are in the particulate form and 31.2% are  
264 dissolved  $\text{PO}_4\text{-P}$ .

265 Concerning the daily fluctuations,  $\text{NH}_4\text{-N}$  flux profiles (figure 6) show higher values during  
266 the day and decrease in the night. In spite of a slight  $\text{NH}_4\text{-N}$  increase 4 to 6 hours after the  
267 morning feed distribution, the two daily feed distributions seem to reduce the postprandial  
268 excretion peak. SS daily fluxes show higher fluctuations (figure 7). There is a time lag  
269 between  $\text{NH}_4\text{-N}$  and SS fluxes: SS transit seems to be slower than excretion. The  
270 concentrations of other substances are lower and more stable during the day.

271

## 272 **Discussion and conclusion**

273 The CEMAGREF method gives lower SS value than the nutritional method and the measured  
274 value (Table III and figure 2). This can be explained by excessive variation coefficients of the  
275 results of this model, which is not statistically acceptable for the SS (Jatteau, 1999), and by  
276 important daily SS fluctuations (figure 7). The predicted daily flux of total-P calculated using  
277 the nutritional method is quite similar to the CEMAGREF estimation and lower than the  
278 measured value. The  $\text{NH}_4\text{-N}$  fluxes calculated with the three methods are in the same order of  
279 magnitude. Even if the CEMAGREF method gives consistent results, this method is only  
280 based on the daily quantity of feed distributed and do not take into account the feed  
281 composition or the digestibility coefficients, while they are currently drastically improved. In  
282 fact, metabolic wastes can be minimised by modifying the digestibility, the energetic density  
283 and friability of the feed ingredients (Cho and Bureau, 1997; Kaushik, 1998; Roque  
284 d'Orbcastel and Blancheton, in press, 2006). MacMillan *et al.* (2003) attributed 40% of the P  
285 effluent reduction of flow-through trout farms, during the past 15 years, to management  
286 improvements, such as feeding practices, low-P (0.9%) feed use and frequent tank cleanings  
287 (quiescent zone management).

288

289 In our study, the total annual waste production estimated with the nutritional method,  
290 expressed per metric ton of fish standing stock, were 147.5 kg for solids, 40.8 kg for N, and  
291 8.7 kg for P, lower than those reported by Axler *et al.* (1997) and by Bureau *et al.* (2003) for  
292 salmonid farms (table IV).

293 Concerning the comparison between the nutritional method and the hydrobiological method  
294 results, predicted and measured N waste fluxes are quite similar: the predictions and  
295 measurements are well correlated ( $r^2 = 0.88$ ), with predictions a bit higher than measurements.  
296 For the TP and SS parameters, the predicted and measured fluxes are less correlated ( $r^2$  of  
297 0.53 and 0.48 respectively), with measurements higher than predictions. The physical  
298 properties of solid wastes, subject to decantation as well as re-suspension, can explain part of  
299 the differences. According to Boujard *et al.* (1999) and Papatryphon *et al.* (2005), N, P and  
300 SS are sometimes underestimated by the hydrobiological method because of sampling  
301 difficulties and sample preservation difficulties, and sometimes overestimated, because of  
302 solid re-suspension (due to fishing, tank cleaning or hydrology). They can also be under or  
303 overestimated by the nutritional method, depending on the digestibility coefficients and the  
304 precision of ingested feed quantities.

305 Boujard *et al.* (1999) compared the results of waste evaluation with the nutritional and the  
306 hydrobiological methods (two consecutive 24 h periods, with samples taken every 2 hours, on  
307 4 rainbow trout tanks). They found a global balance of nitrogenous wastes of 50-65 g N.kg  
308 feed<sup>-1</sup> and 9-16 g P.kg feed<sup>-1</sup> for the phosphorous corresponding value, a bit higher than those  
309 found during the present study. In their study, they defined the waste as the fraction of the  
310 nutrients which are not retained by the fish, including also the uneaten feed (Boujard, *pers.*  
311 *comm.*). The lower quantities that we measured using the hydrobiological method ( $38.5 \pm 7.1$   
312 of total-N g.kg<sup>-1</sup>feed and  $9.7 \pm 2.5$  of total-P), could be explained by better feed management  
313 on the Murgat farm which results in almost no uneaten feed. They shown also a good

314 correlation between predicted and measured N values, with  $r^2 = 0.85$ , higher than the  
315 correlation factor for P values of 0.67. According to the authors, the wastes measured with the  
316 hydrobiological method were underestimated but comparable to the calculated values. They  
317 attributed this underestimation to the settleable characteristic of the suspended solids.

318 Papatryphon *et al.* (2005) compared the predicted values with  $\text{NH}_4^+$ , TP and SS  
319 concentrations measurements in the recipient river. They found waste prediction values well  
320 correlated with the measured values, but the trend was an overestimation of predicted  $\text{NH}_4^+$   
321 and P values, that the authors explained by a probable degradation of  $\text{NH}_4^+$  in the samples  
322 through nitrification processes. Some observed concentrations in SS were higher than  
323 predictions, certainly due to the highly variable solid transport in aquaculture raceways (solids  
324 decantation or re-suspension), which depends on the farm management and/or environmental  
325 variability such as high flow rate. Maillard *et al.* (2005) observed higher TSS concentrations  
326 during harvesting and feeding events (fish agitation) of different raceway system trout farms.

327

328 Both methods present drawbacks and advantages. The hydrobiological method is interesting  
329 because it gives details on the different forms of N and P in the wastes (Boujard *et al.*, 1999),.  
330 The results obtained in this study are comparable to those of previous studies: (Braaten, 1991;  
331 Heinen *et al.*, 1996; True *et al.*, 2004) reported that over 85% of N was in dissolved form and  
332 40-85% of P in solid form. Boujard *et al.* (1999) found that for 1 kg of dry feed (80-93 g of N  
333 and 12-21 g of P) similar results for the N waste proportions (73% of the nitrogen was  
334 released, with 78% in  $\text{NH}_4\text{-N}$  form) but opposite for the P wastes (87% of the phosphorous  
335 was released with 60% in dissolved form (mainly  $\text{PO}_4\text{-P}$ )).

336 Using the hydrobiological method, we observed important daily  $\text{NH}_4\text{-N}$  and SS fluxes  
337 fluctuations (figure 8). In fact, fish farm wastes are highly fluctuating: daily variations  
338 depending on feeding time and farm management (fishing, sorting...); annual variations

339 depending on the fish biomass and distributed feed. For example,  $\text{NH}_4$  waste increases after  
340 the feeding time, with a maximum around 6 hours after feeding, depending on species, feed  
341 and feeding ratio and feeding several times a day contributes to decrease the waste daily  
342 fluctuation (Dosdat *et al.*, 1996; Jatteau, 1999). SS fluxes increase during the feeding period  
343 because of fish motion and may also increase after digestion (after Guillaume *et al.*, 1999,  
344 ingested feed stays in the gut of 250-500g fishes during about 10 hours after ingestion).

345 Representative samples of the waste produced by the farm cannot be obtained if the number  
346 of samples is decreased (Boujard *et al.*, 1999; Cho and Bureau, 1997; Jatteau, 1999). Several  
347 sampling periods have to be implemented simultaneously in the inlet and outlet of the farm in  
348 order to get representative results. Sampling must be done carefully, especially because of the  
349 solid matter properties. The AFNOR-NFT90-105 recommends a sample of a minimum  
350 volume of 500ml (for fresh water). The samples have to be preserved because of the  
351 possibility of nutrient transformation through leaching and bacterial activity.

352 For the hydrobiological method, the main difficulty is the water flow rate measurement, a key  
353 point for the flux evaluation but difficult even with a precision equipment. From one tank to  
354 another, even if the geometry is the same, the measured flow rate varies by 20%. From one  
355 day to another, the variation of the flow rate measurement could be around 35%. The  
356 difficulty in evaluating the water flow rate makes current waste control validity questionable.

357 Environmental monitoring is based on the use of indicators, such as the maximum SS, BOD,  
358  $\text{NH}_4$  concentrations in the recipient ecosystem. As fluxes are calculated with concentration  
359 and flow rate, it seems to be difficult to properly control the correlation between the measured  
360 and the predicted values at the farm outlet (with their own uncertainties) as recorded by the  
361 farmer in the environmental assessment.

362 The hydrobiological method appears to be too heavy and costly for regular use as part of the  
363 waste quantification and self monitoring processes required under the ICPE legislation.

364 In comparison, the nutritional method is easier and quicker, and a rather inexpensive way to  
365 predict fish waste production. Using the theoretical digestibility coefficients (Papatyphon *et*  
366 *al.*, 2005) and feed composition given by the manufacturer, or the measured digestibility  
367 coefficients (for proteins and lipids) and feed composition, the nutritional method gave  
368 different solid waste evaluation. With the theoretical protein, lipid and carbohydrate  
369 coefficients and theoretical feed composition, the SS predicted emissions are 88.5 tons / year  
370 whereas with measured coefficients, the model gives 69.3 tons / year. So the feed composition  
371 and the digestibility coefficients used in the model can lead to more than 20% variation in the  
372 solid waste evaluation.

373 Even if the hydrobiological and nutritional methods do not allow one to precisely anticipate  
374 waste production, both provide interesting orders of magnitude; the nutritional method is the  
375 simplest for the fish farmers to evaluate the waste produced by their farm, although it requires  
376 precise information (especially on feed composition, ingested feed quantity and digestibility  
377 coefficients are available).

378

379 If it is established that waste emissions can be reduced at the fish level (Cho and Bureau,  
380 1997; Kaushik, 1998; Roque d'Orbcastel and Blancheton, 2006; MacMillan *et al.* 2003),  
381 waste also has to be reduced at the system level through the use of well designed waste  
382 treatment systems. The design of the treatment systems also requires good knowledge of the  
383 waste production process especially because the economic feasibility of aquaculture waste  
384 treatment has not yet been demonstrated in most of the situations.

385

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## References

- Axler, R.P., Tikkanen, C., Henneck, J., Schuldt, J., and McDonald, M.E., 1997. Characteristics of effluent and sludge from two commercial rainbow trout farms in Minnesota. *The Progressive Fish Culturist*, 59, 161-172.
- Benschneider, K., and Robinson, R.J., 1952. A new spectrophotometric determination of nitrite in seawater. *J. Mar. Res.*, 11, 87-96.
- Bergheim, A., and Brinker, A., 2003. Effluent treatment for flow through systems and European environmental regulations. *Aquac. Eng.*, 27, 61-77.
- Boujard, T., Vallée, F., and Vachot, C., 1999. Evaluation des rejets d'origine nutritionnelle de truiticultures par la méthode des bilans, comparaison avec les flux sortants. *Dossier de l'environnement*, INRA (Eds), 26, 32-35.
- Braaten, B., 1991. Impact of pollution from aquaculture in six Nordic countries. Release of nutrients, effects, and wastewater treatment. In: De Pauw, N., Joyce, J. N. (Eds), *Aquaculture and the Environment*. European Aquaculture Society Publication 16, Ghent, Belgium, 79-101.
- Bureau, D.P., Gunther, S.J., Cho, C.Y., 2003. Chemical composition and preliminary theoretical estimates of waste outputs of rainbow trout reared in commercial cage culture operations in Ontario. *N. Am. J. Aquacult.*, 65, 33-38.
- Cho, C.Y., 1993. Digestibility of feedstuffs as a major factor in aquaculture waste management. In: *Fish Nutrition in Practice*, S.J., Kaushik and P., Luquet (Eds), INRA, Paris, 61, 365-374.
- Cho, C.Y., and Bureau, D.P., 1997. Reduction of waste output from salmonid aquaculture through feeds and feedings. *The Progressive Fish Culturist*, 59, 155-160.

Cho, C.Y., and Bureau, D.P., 1998. Development of bioenergetic models and the Fish-PrFEQ software to estimate production, feeding ration and waste output in aquaculture. IN Proc. of the 3rd International Symposium on Nutritional Strategies and Management of Aquaculture Waste. *Aquat. Living Resour.*, 11, 199-210.

Cho, C.Y., Hynes, J.D., Wood, K.R., and Yoshida, H.K., 1991. Quantification of fish culture wastes by biological (nutritional) and chemical (limnological) methods; the development of high nutrient dense (HND) diets. *Nutritional Strategies and Aquaculture Waste. Proceedings of the 1st International Symposium on Nutritional Strategies in Management of Aquaculture Waste*, Guelph, Ontario, Canada. *In: Cowey, C.B., and Cho, C.Y., (Eds).*

Cho, C.Y., Hynes, J.D., Wood, K.R., and Yoshida, H.K., 1994. Development of high nutrient-dense, low pollution diets and prediction of aquaculture wastes using biological approaches. *Aquaculture*, 124, 293-305.

Company, R., Caldach-giner, J.A., Perez-sanchez, J., and Kaushik, S.J., 1999. Protein sparing effect of lipids in common dentex (*Dentex Dentex*): A comparative study with sea bream (*Sparus aurata*) and sea bass (*Dicentrarchus Labrax*). *Aquat. Living Resour.*, 12, 23-30.

Dosdat, A., 1992a. L'excrétion chez les poissons téléostéens I. Azote. *Piscic. Fr.*, 108, 25-37.

Dosdat, A., 1992b. L'excrétion chez les poissons téléostéens II. Le Phosphore. *Piscic. Fr.*, 109, 18-28.

Dosdat, A., Servais, F., Métailler, R., Huelvan, C., and Desbruyères, E., 1996. Comparison of nitrogenous losses in five teleost fish species. *Aquaculture*, 141, 107-127.

Dosdat, A., 1999. Rappels de physiologie et de métabolisme. *In: Aquaculture et environnement, tome 1, aspects techniques et économiques*, Jean Petit (ed.). INRA, Paris, 69-74.

Fauré, A., 1983. *Salmoniculture et Environnement*, Vol 1. Evaluation de la pollution rejetée par les salmonicultures intensives. CEMAGREF, Bordeaux, France, Etude n°16, 71 pp.

Fernandes, T.F., Miller, K.L., and Reard, P.A., 2000. Monitoring and regulation of marine aquaculture in Europe. *J. Appl. Ichthyol.*, 16, 138-143.

Guillaume, J., Kaushik, S., Bergot, P., and Métailler, R., 1999. Bases de la nutrition des animaux aquatiques : physiologie digestive et digestibilité des nutriments. *Nutrition et alimentation des poissons et crustacés*. Éditions INRA-IFREMER, 489 pp, 71-75.

Heinen, J.M., Hankins, J.A., and Adler, P.R., 1996. Water quality and waste production in a recirculating trout-culture system with feeding of a higher energy or a lower energy diet. *Aquac. Res.*, 27, 699-710.

Hennessy, M.M., Wilson, L., Struthers, W., and Kelly, L. A., 1996. Waste loadings from two freshwater atlantic salmon juvenile farms in Scotland. *Water , Air and soil pollution*, 86, 235-49.

Huhta, C., and Ward, C.J., 2003. Flow Measurements using an Upward-Looking Argonaut-SW Doppler Current Meter: *in IEEE/OES Seventh Working Conference on Current Measurement Technology*, San Diego, CA, March 10-13, 2003, Proceedings.

Jatteau, P., 1999. Quantification des flux polluants. *Aquaculture et environnement*, tome 1, aspects techniques et économiques, Jean Petit (éd.). INRA, Paris, pp 74-87.

Kaushik, S.J., 1980. Influence of nutritional status on the daily patterns of nitrogen excretion in the carp (*Cyprinus carpio L.*) and the rainbow trout (*Salmo gairdneri R.*). *Reprod. Nutr. Develop.*, 20 (6), p 1751.

Kaushik, S.J., 1998 . Nutritional bioenergetics and estimation of waste production in non salmonids. *Aquat. Living Resour.*, 11, 211-217.

- Kelly, L.A., Bergheim, A., and Hennessy, M.M., 1994. Predicting output of ammonium from fish farms. *Wat. Res.*, 28 (6), pp 1403-1405.
- Lemarié, G., Martin, J.L.M, Dutto, G., and Garidou, C., 1998. Nitrogenous and phosphorous waste production in a flow-through land-based farm of European seabass (*Dicentrarchus labrax*). *Aquat. Living Resour.*, 11, 247-254.
- Liao, P., 1970. Pollution potential of salmonids fish hatcheries. *Water sewage works*, 117, 291-297.
- Liao, P., and Mayo, R., 1974. Intensified fish culture combining water reconditioning with pollution abatement. *Aquaculture*, 3, 61-85.
- MacMillan, J.R., Huddleston, T., Woolley, M., and Fothergill, K., 2003. Best management practice development to minimize environmental impact from large flow-through trout farms. *Aquaculture*, 226, 91-99.
- Maillard, V.M., Boardman, G.D., Nyland, J.E., and Kuhn, D.D., 2005. Water quality and sludge characterisation at raceway-system trout farms. *Aquac. Eng.*, 33, 271-284.
- Maroni, K., 2000. Monitoring and regulation of marine aquaculture in Norway. *J. Appl. Ichthyol.*, 16, 192-195
- Papatryphon, E., Petit, J., Hayo, V., Kaushik, S.J., and Claver, K., 2005. Nutrient-balance modelling as a tool for environmental management in aquaculture: The case of trout farming in France. *Env. Managem.*, 35 (2), 161-174.
- Pedersen, P.B., 1999. Monitoring and regulation of marine aquaculture in Denmark. *J. Appl. Ichthyol.*, 16, 144-147.

Roque d'orbcastel E., and Blancheton, J.P., 2006. The wastes from marine fish production systems : characterization, minimization, treatment and valorization. *World aquaculture*, 37, 3, 28-35, 70.

Solorzano, L., 1969. Determination of ammonia in natural waters by the phenol-hypochlorite method. *Limnol. Oceanogr.*, 14, 799-801.

Tarazona, J.V., Ortiz, J.A., Carbello, M., and Munoz, M.J., 1993. Pollution generated by fish farms. A systems dynamics model. *Fresenius Environmental Bulletin*, 2, 84-89.

True, B., Johnson, W., and Chen, S., 2004. Reducing phosphorous discharge from flow-through aquaculture I: facility and effluent characterisation. *Aquac. Eng.*, 32, 129-144.

Wood, E.D., Armstrong, F.A.J., and Richards, F.A., 1967. Determination of nitrate in sea water by cadmium cooper reduction to nitrite. *J. Mar. Biol.*, 47, 23-31

Table I. fish extruded feed composition (%), theoretical nutrient digestibility coefficients (DC) (from Papatryphon *et al.*, 2005) and calculated digestibility coefficients (%) (Moutounet, *pers. comm.*)

	Mean feed composition (%)	Theoretical DC (%)	Calculated DC (%)
Moisture	8		
Protein	45	90	93
Lipids	27	95	96
Carbohydrate	10.1	60	75
Ash	6.7	50	
Fibre	1.4	0	
Phosphorus	0.9	50	
Energy (MJ.kg <sup>-1</sup> )	21.2		

Table II. Biomass in tanks, daily feed quantities, average feeding rates and FCR of the farm during the different sampling series (last serie only includes the sector 1 results; sector 2 was not sampled because of too important fishing events)

Date	Biomass (kg)	Daily feed (kg.d <sup>-1</sup> )	Average feeding Rate (%)	Average FCR (kg.kg <sup>-1</sup> )
25-26.01.2006	177 449	1314	0.74	0.88
07-08.02.2006	174 412	1333	0.76	0.87
22-23.02.2006	178 571	1568	0.88	0.88
07-08.03.2006	130 643	1012	0.84	0.77



Table III. Daily waste production of the whole farm, predicted according to the nutritional method and measured in situ with the hydrobiological method, expressed in  $\text{kg}\cdot\text{d}^{-1}$  and  $\text{g}\cdot\text{kg}^{-1}$  feed delivered.  $\text{d}^{-1}$

Parameter	Measured mean fluxes ( $\text{kg}\cdot\text{d}^{-1}\pm\text{S.D.}$ )	Predicted mean fluxes ( $\text{kg}\cdot\text{d}^{-1}\pm\text{S.D.}$ )	Cemagref calculated values ( $\text{kg}\cdot\text{d}^{-1}\pm\text{S.D.}$ )	Measured mean fluxes ( $\text{g}\cdot\text{kg}^{-1}\text{ feed}\cdot\text{d}^{-1}\pm\text{S.D.}$ )	Predicted mean fluxes ( $\text{g}\cdot\text{kg}^{-1}\text{ feed}\cdot\text{d}^{-1}\pm\text{S.D.}$ )
Suspended solids	$317.8 \pm 165.7$	$206.5 \pm 20.7$	$136.3 \pm 14.1$	$226.2 \pm 117.9$	$147.0 \pm 0.2$
Total nitrogen	$54.1 \pm 10$	$59.8 \pm 6.0$		$38.5 \pm 7.1$	$42.6 \pm 0.4$
Particulate nitrogen	$11.8 \pm 3.4$	$10.1 \pm 1.0$		$8.4 \pm 2.4$	$7.2 \pm 0.0$
Ammonia nitrogen	$31.6 \pm 7.5$	$39.7 \pm 4.0$	$36.4 \pm 3.7$	$22.5 \pm 5.3$	$28.3 \pm 0.3$
Urea nitrogen	$10.7 \pm 2.5$	-		$7.6 \pm 1.8$	-
Total phosphorus	$13.6 \pm 3.5$	$6.3 \pm 0.6$	$6.7 \pm 0.7$	$9.7 \pm 2.5$	$4.5 \pm 0.1$
Particulate phosphorus	$9.6 \pm 3.6$	-		$6.8 \pm 2.6$	-
Orthophosphate-P	$4.0 \pm 0.2$	-		$2.8 \pm 0.1$	-

Table IV. Total annual waste production of the farm calculated with the nutritional method, in comparison with values reported by Axler *et al.* (1997) and Bureau *et al.* (2003), expressed per metric ton of fish

Parameter	Calculated values (kg. ton <sup>-1</sup> of fish produced )	Axler <i>et al.</i> (1997) values (kg. ton <sup>-1</sup> of fish produced )	Bureau <i>et al.</i> (2003) values (kg. ton <sup>-1</sup> of fish produced )
Suspended solids	147.5	289-839	240-318
Total nitrogen	40.8	47-87	47-71
Total phosphorus	8.7	4.8-18.7	7.5-15.2

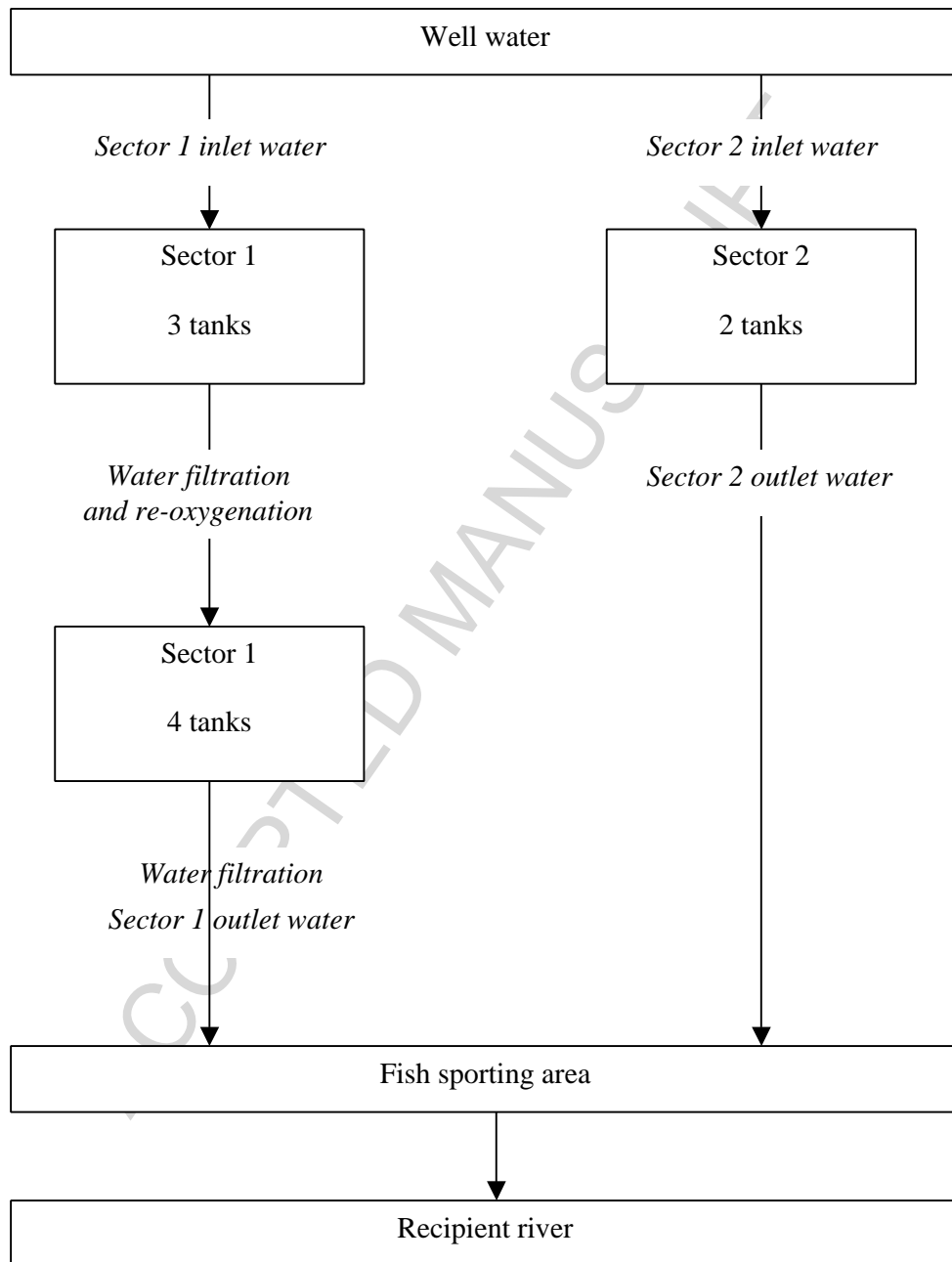
**Figure captions**

Figure 1. The growing sector of the farm, divided into two sectors: sector 1 composed of 7 concrete tanks with 4 species reared and sector 2 composed of 2 concrete tanks with only rainbow trout species. Each sector is fed by its own well water.

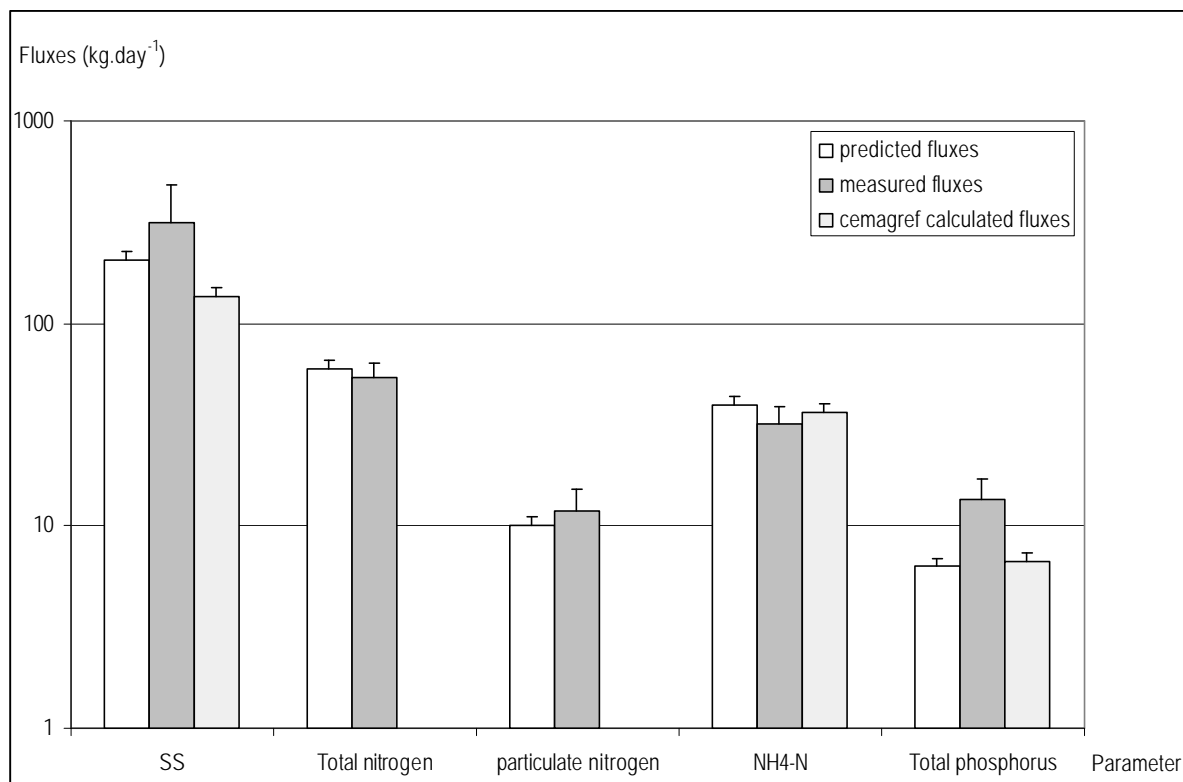


Figure 2. Predicted (nutritional method and CEMAGREF method) and measured fluxes of the farm, expressed in kg per day, with a logarithmic scale.

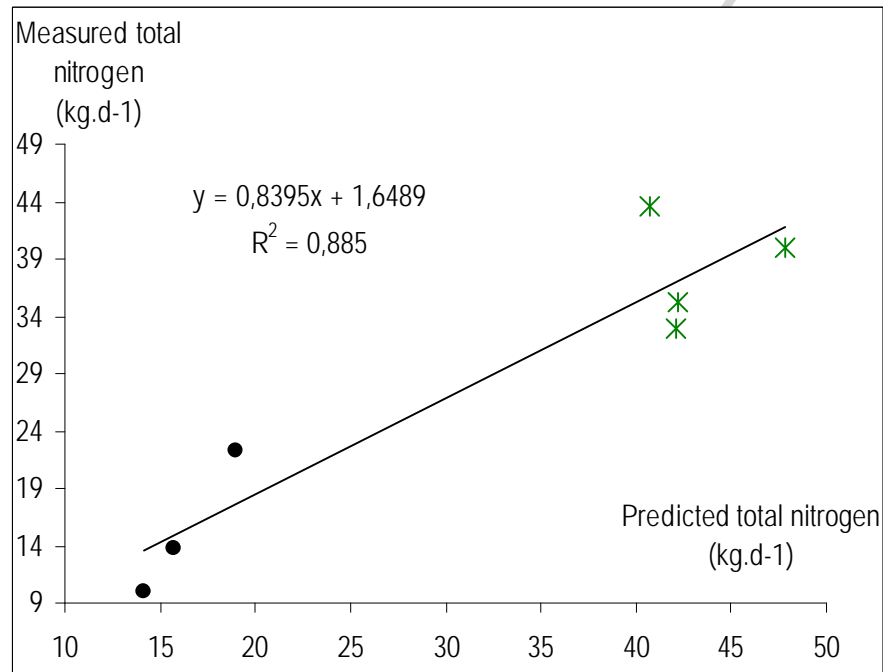


Figure 3. Comparison of the total-N measured values and the total-N predicted values, in the two different areas of the farm (sector 1 values are represented with green stars, sector 2 values with black points). The measured values are obtained from the hydrobiological method, the predicted values from the nutritional method. Total-N is the total-N flux produced by the farm during a day, expressed in kg per day.  $R^2$  is the correlation factor.

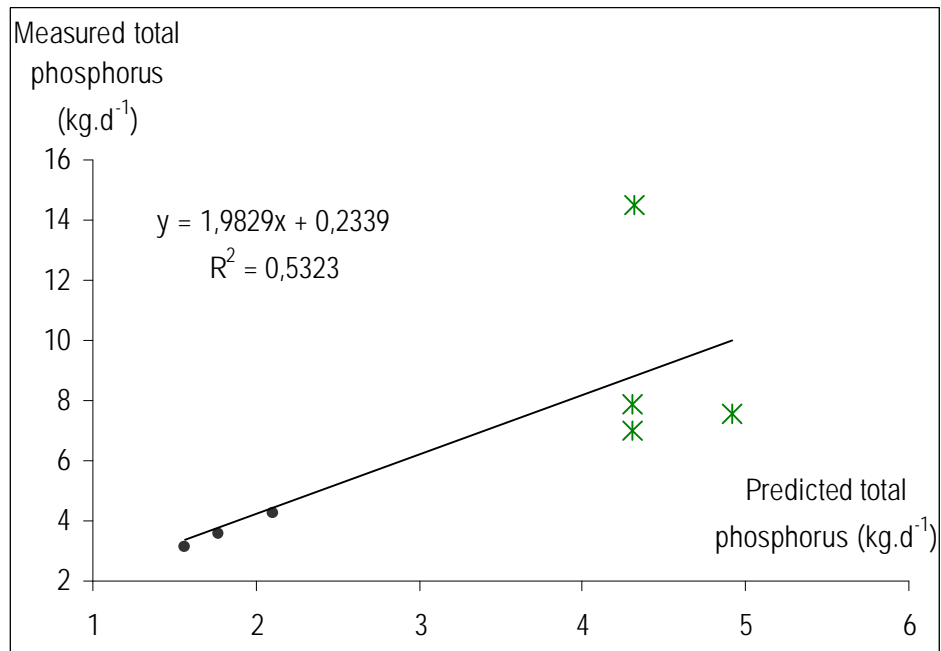


Figure 4. Comparison of the total-P measured values and the total-P predicted values, in the two different areas of the farm (sector 1 values are represented with green stars, sector 2 values with black points). The measured values are obtained with the hydrobiological method, the predicted values with the nutritional method. Total-P is the total-P flux produced by the farm during a day, expressed in kg per day.  $R^2$  is the correlation factor.

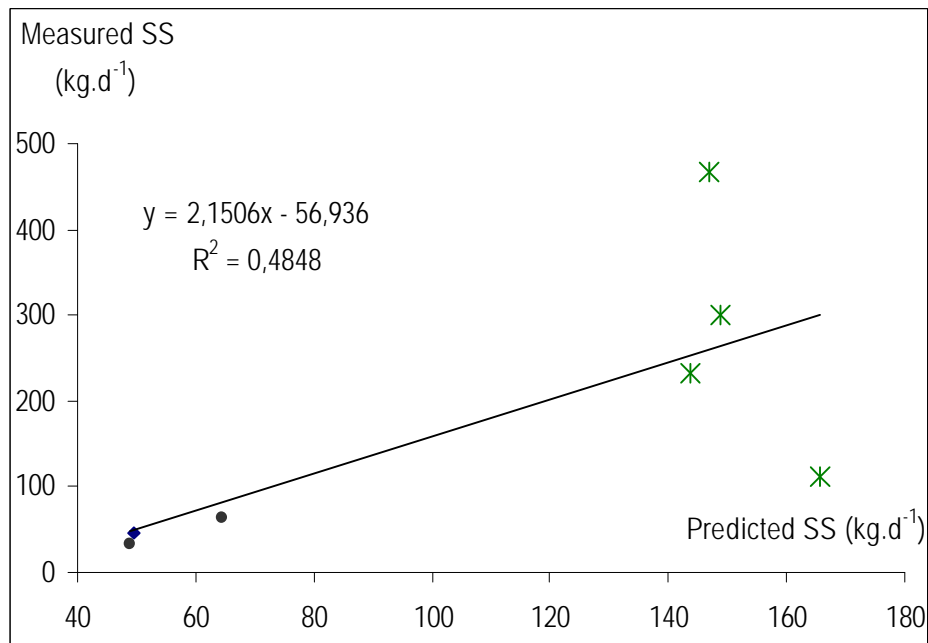


Figure 5. Comparison of the total suspended solid measured values and the total suspended solid predicted values, in the two different areas of the farm (sector 1 values are represented with green stars, sector 2 values with black points). The measured values are obtained with the hydrobiological method, the predicted values with the nutritional method. TSS is the total suspended solid flux produced by the farm during a day, expressed in kg per day.  $R^2$  is the correlation factor.

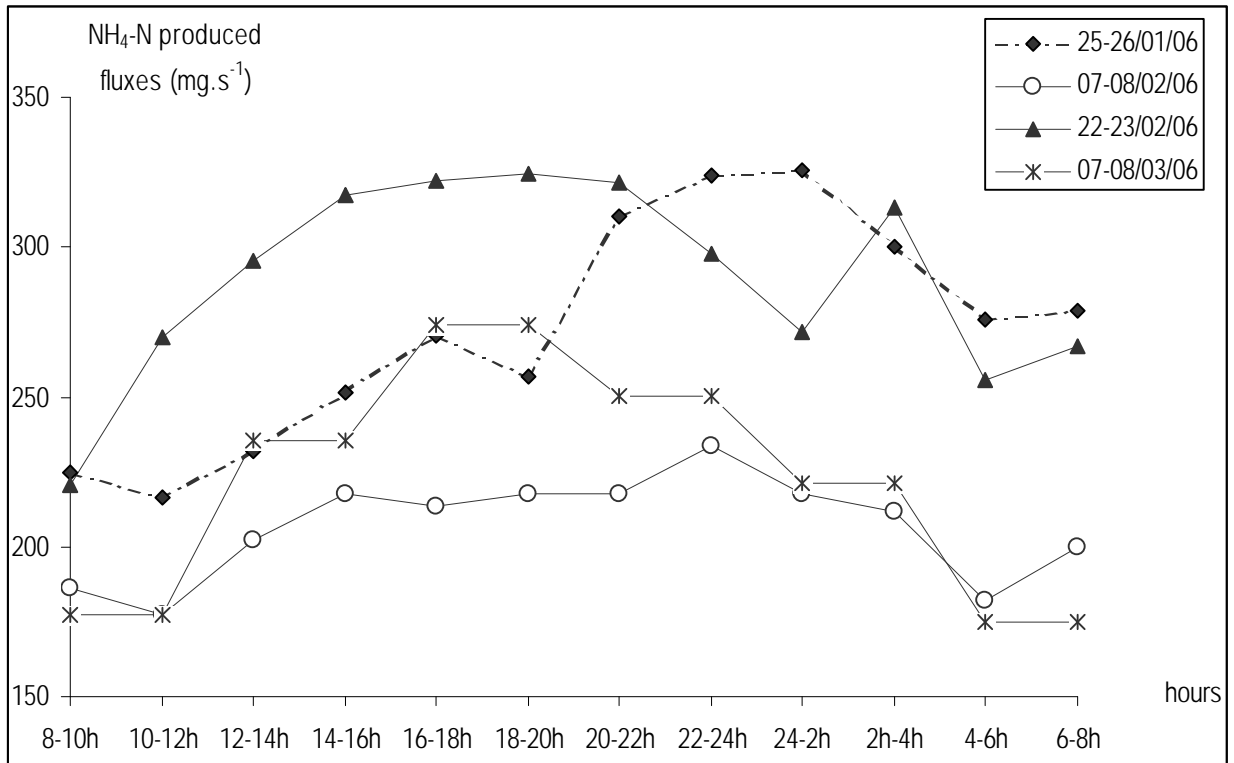


Figure 6. Daily fluctuations of the  $\text{NH}_4\text{-N}$  produced by the farm (sectors 1 & 2), for 3 different 24h sampling periods (1: 25-26.01.2006; 2: 07-08.02.2006; 3: 22-23.02.2006) and produced by the sector 1 only for the last date (06-07.03.06). The  $\text{NH}_4\text{-N}$  fluxes are expressed in mg per second.



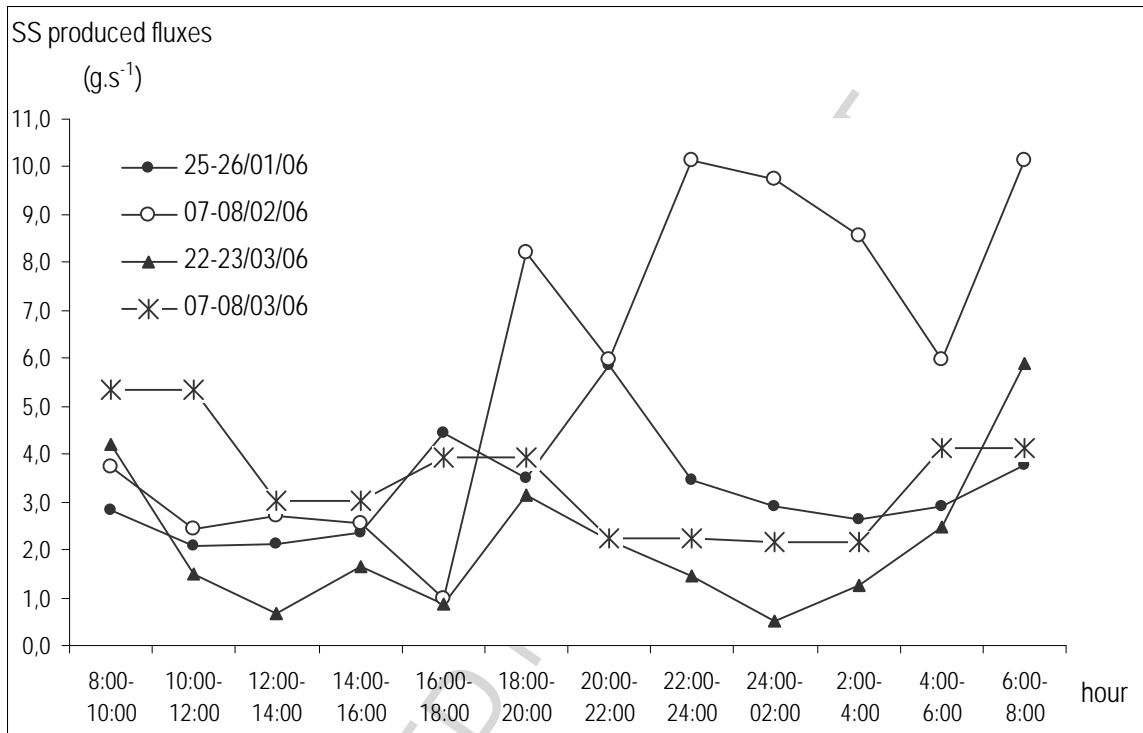


Figure 7. Daily fluctuations of the total suspended solids produced by the farm, for the first 3 24h sampling periods (1: 25-26.01.2006; 2: 07-08.02.2006; 3: 22-23.02.2006) and produced by the sector 1 for the last date (06-07.03.06). The TSS fluxes are expressed in g per second.

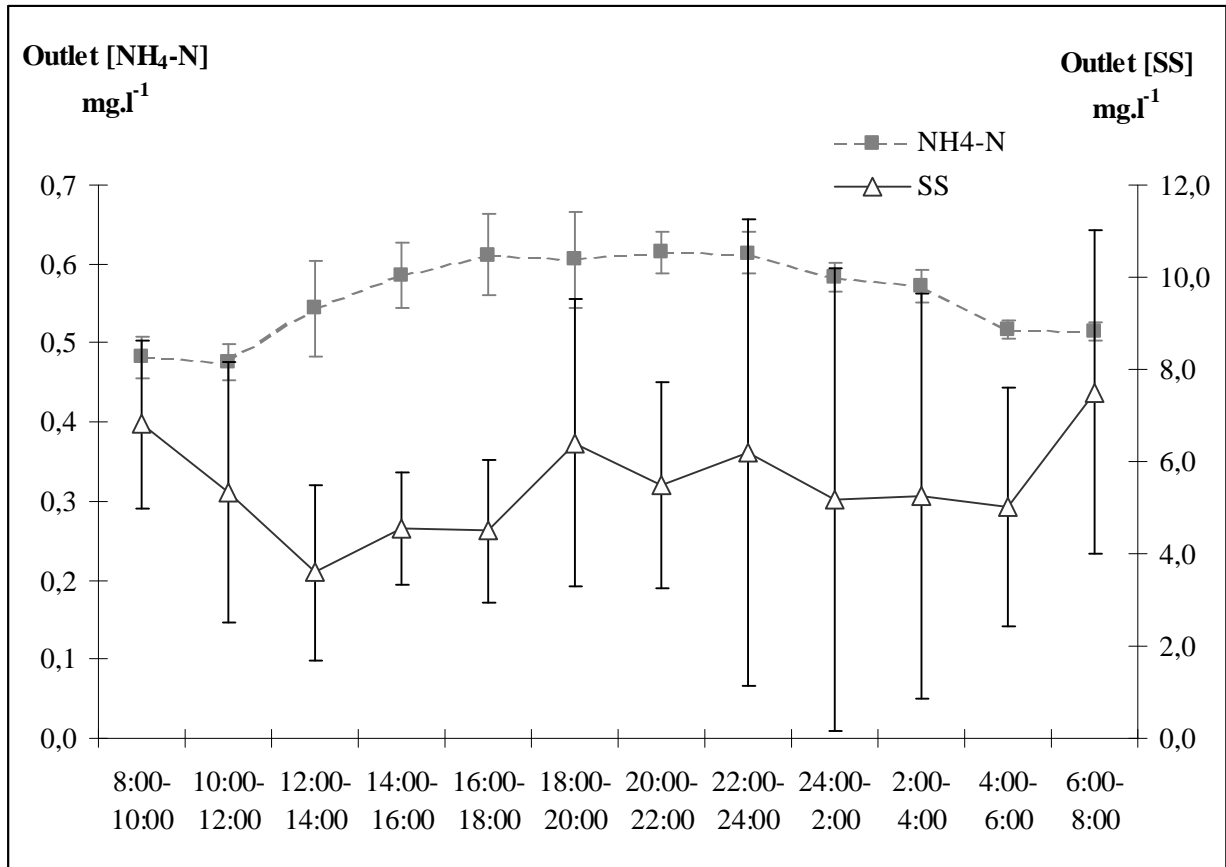


Figure 8. Averaged suspended solid and NH<sub>4</sub>-N outlet concentrations (with standard deviations), measured at the outlet point of the farm, during four different 24h sampling periods. The concentrations are expressed in mg per litre.

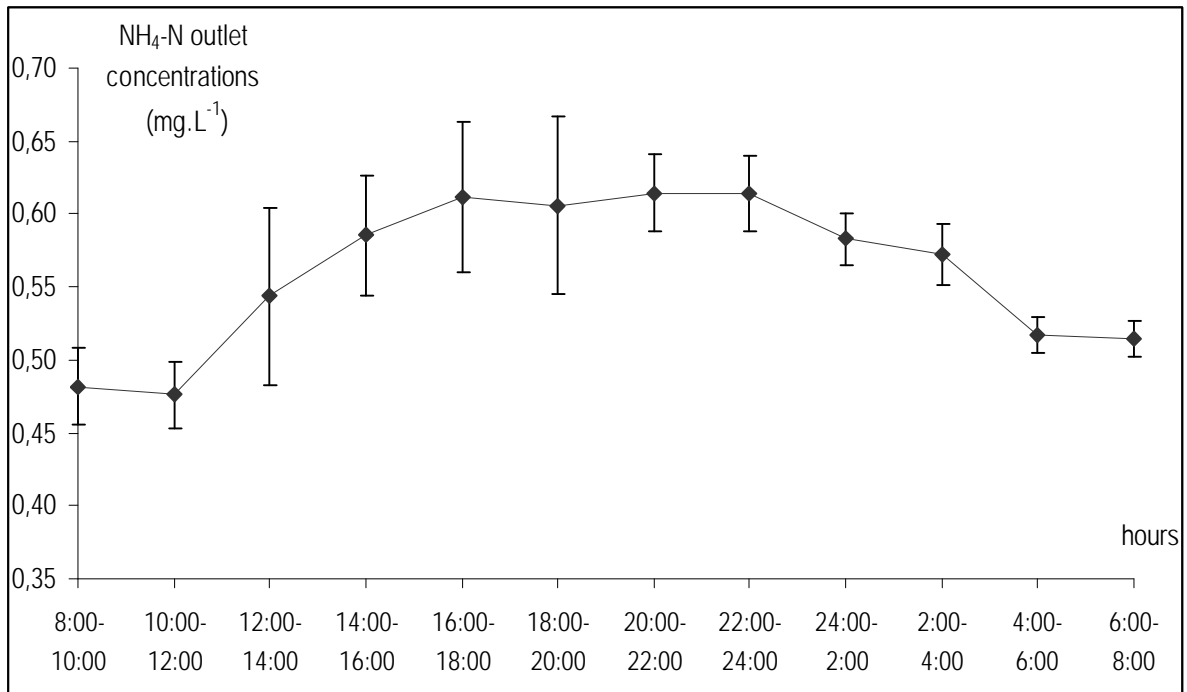


Figure 9. Averaged  $\text{NH}_4\text{-N}$  outlet concentrations (with standard deviations), measured at the outlet point of the farm, during four different 24h sampling periods. The concentrations are expressed in mg per litre.